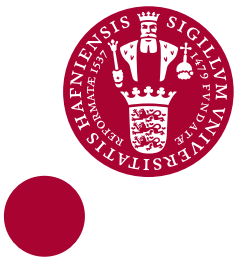

Evidence of TRA-1-60 and TRA-1-81 involvement in L-selectin mediated adhesion of the porcien embryo

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[P09.16].
EVIDENCE OF TRA-1-60 AND TRA-1-81 INVOLVEMENT IN L-SELECTIN MEDIATED ADHESION OF THE PORCINE EMBRYO

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L-selectin expression in the early human embryo is involved in trophoblast adhesion during implantation. The aim of this study was to investigate the potential role of the L-selectin adhesion-system in the adhesion and placentation of porcine embryos. Endometrial samples were collected from pregnant gilts and non-pregnant cycling sows at Days 10, 15 and 18 post insemination/oestrus. Embryos were collected from pregnant gilts at Days 9, 11 and 15 for expression studies and from Days 10 and 15 for immunohistochemistry.
Expression analysis of L-selectin and its ligands was made using qPCR. Protein localization of L-selectin and the ligands PNAd, PEN5 as well as the podocalyxin epitopes Tra-1-60 and Tra-1-81 were investigated by immunohistochemistry. In pregnant gilts the mRNA expression of L-selectin was approximately 14 fold up-regulated at Days 15 and 18 compared to Day 10 p.i. No significant changes were seen in the mRNA levels in the cyclic sows. Expression of mRNA for the potential L-selectin ligands, CSPG-2 and podocalyxin were 6 and 12 times up-regulated in tubular Day 11 blastocysts compared to Day 9 blastocysts. Staining for L-selectin was observed in the luminal epithelium of the endometrium. In the embryos, staining for MECA-79 and PEN5 were observed in trophoblast and hypoblast cells at day 15. Tra-1-81 and Tra-1-60 was confined to the epiblast at Day 10 but at Day 15 staining for both epitopes were observed in the trophoblast. Our results for the first time demonstrate that key components of the L-selectin adhesion system are present in the porcine uterus and embryo around the time of initial placentation. Interestingly, the components are expressed in a pattern opposite to that found in man: In the pig, L-selectin is expressed in the endometrium and the ligands in the trophoblast with the reverse being true in man.
Keywords: Porcine, Implantation, L-selectin

[P10.01].
EFFECTS OF INSULIN ON MEMBRANE LIPID COMPOSITION OF HUMAN SYNCYTIOTROPHOBLAST

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Introduction: Altered placental membrane lipid composition in pregnancy may affect the fetal-maternal exchange. With gestational progress, the composition, structure and functions of these membrane lipid bilayers are modified in order to meet the changing metabolic needs of the growing fetus.
Previously, we reported that plasma membranes of syncytiotrophoblast (hST) are unusual in comparison to other cell types. Because of the increase of sphingomyelin we also informed that preeclamptic hST is more rigid than normal hST. Since we observed high serum levels of insulin in preeclamptic women, we hypothesized that insulin may be implicated in the changes observed in preeclampic hST.
The aim of this study was to evaluate if the insulin may alter the lipid composition of hST.
Methods: Explants from normal term placentas were cultured with different concentrations of insulin during 24 h. The biochemical viability of the explants was determined by estimation of β-hCG concentrations in the extracellular medium.
Apical (MVM) and basal (BM) membrane vesicles were prepared by differential centrifugation.
Lipid were extracted by Bligh-Dyer method and quantified by Fiske-Subarrow. Cholesterol was determined by enzymatic method.
Results: Insulin treatment produced no changes on the total phospholipid concentration in BM. On the contrary, MVM showed an increase in total lipid content until reaching a constant value at 100 μUI/mL of insulin.
There were no changes in the amount of cholesterol in both vesicles.
Discussion: Our results suggest that insulin treatment only alter phospholipid concentration in MVM having no effect on BM vesicles. Further work is necessary to clarify the molecular mechanisms implicated in these changes and if they may play a role in the pathogenesis of preeclampsia.
Keywords: insulin, lipids, syncytiotrophoblast, preeclampsia